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ON THE NEUROSECRETORY SYSTEM OF *RIVULOGAMMARUS SYRIACUS* CHEVREUX

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Studies on neurosecretion in Crustacea were initiated by Perkins (1928) and Koller (1928). Later, Hanstrom's discovery (1931) of the eyestalk hormone in the sinus gland of brachyurans encouraged Brown (1944); Passano (1951); Bliss and Welsh (1952) and others to establish a neurosecretory sinus gland system in crustacea similar to the intercerebralis-cardiacum-allatum system of the insects and to the hypothalamic-hypophyseal system of the vertebrates. Since then, several morphological studies have been made on the neurosecretory system of decapod crustaceans. The significance of the role of the neurosecretory activities in the physiology of these animals has already been emphasized by several workers (Scharrer, 1955; Welsh, 1955; Knowles and Carlisle, 1956).

In Isopoda, study of protocephalic neurosecretory pathway and the corresponding endocrine glands revealed a pleomorphism greater than that observed in any other malacostracan crustaceans (Hanstrom, 1928, 1929; Graber, 1933; Walker, 1936; Stahl, 1938; de Lattin, 1939; Amar, 1950, 1951, 1953; Gabe, 1952; Miyawaki, 1958; Matsumoto, 1959 and Oguro, 1959a, 1959b, 1960, 1961). It is believed that the great structural variations are associated with the systematic position of the animals investigated. There is little information on the protocephalic neurosecretory pathway in Amphipoda and therefore, neurosecretory cells would not appear to have been investigated in this group except the identification of sinus gland in *Gammarus pulex; G. fluviatilis* and *G. locusta* (Graber, 1933; Stahl, 1938) and cephalic statocyst as an X-organ (Dahl, 1963). Being a primitive malacostracan, it would be interesting to compare the protocephalic neurosecretory pathway at one end with higher malacostracans and at the other end with primitive crustaceans. The present paper describes the structure of various types of neurosecretory cells found in the central nervous system of *Rivulogammarus syriacus* Chevreux, the distribution of these cells and the modes of secretion and discharge of the neurosecretory material in them.

Material and Methods

The freshwater *Rivulogammarus syriacus* was collected from the running water of Nowran in Mosul, Iraq. The average size of the female was 6.5 mm, ranging from 5.0 mm to 10.0 mm and of the male was 7.0 mm ranging from 5.0 mm to 11.0 mm. Whole specimens were fixed in aqueous Bouin, Helly and Carnoy fixatives. The anterior region was decapitated and after dehydration in alcohol and in xylene, was embedded in paraffin. Serial horizontal sections of 8 μ thickness were obtained. The following stainings and histochemical reactions were employed: (1) Bargmann's (1949) modification of Gomori chrome haematoxylin phloxine

technique (CHP), (2) Gabe's (1953) modification of Gomori paraldehyde fuschin technique (PF), (3) Mallory's triple stain, (4) Heidenhain's Azan according to Hubschman (1962), (5) Ehrlich's haematoxylin and eosin (HE).

Rivulogammarus syriacus is a common inhabitant of Nowran stream, found all round the year underneath the stones and sometimes adhered to the algal and other aquatic plants. Their distribution is confined to the areas where the water is shallow and the current is slow. In spring and autumn, the frequency of *Rivulogammarus* is maximum and females and males have been seen in pairs. During these seasons the females carry eggs and juveniles. It indicates that *Rivulogammarus* has two breeding seasons in a year. They are inactive in winter as well as in summer.

Results

Nervous system

The central nervous system of *Rivulogammarus syriacus* consists of a supraesophageal ganglion (brain), circumesophageal commissures, leading to the subesophageal ganglia and a ventral nerve cord. The brain, a vertical mass lies dorsad of the alimentary canal at the anterior surface of the head capsule, bearing dorsolaterally and ventrally two large pair of lobes, is divided into protocerebrum, deutocerebrum and tritocerebrum. These morphological divisions are obscured externally due to the reduction of the deutocerebrum. The protocerebrum is well developed and its dorsolateral lobe tapers towards the optic nerve (Fig. 1). The distal three-fourths of the dorsolateral lobe may be designated as optic lobe, though they are not demarcated by a peduncle as in many decapods. The deutocerebrum is not well developed in comparison with protocerebrum. The tritocerebrum tapers laterally, then posteriorly.

Types of nerve cells and their location

The neurosecretory cells of *Rivulogammarus syriacus* are distinguished into six types on the basis of size, general shape of the cell body, presence or absence of vacuoles in the cytoplasm, and staining properties of the secretory material and have been designated as types: A, B, B′, C, D, and E. (Figs. 2–13) in the present account. These cells exhibit definite localizations and are distributed in several groups in the peripheral region throughout the nervous system. These groups on the dorsal surface of the brain are: medio-dorsal, antero-dorsal, antero-dorso-lateral, dorso-lateral, postero-dorsal, and on the ventral surface are: antero-ventral, ventro-lateral, tritocerebrum, circumesophageal and ventral nerve chain (Fig. 1).

Structure of neurosecretory cells

A cell (*Figs. 2 and 8*). A cells are pyriform and with axons, measure about 15–20 μ in diameter. The cytoplasm has a dense granulated appearance and is full of mitochondria and Nissl's bodies. Minute secretory granules which are stained dark blue with CHP appear in the cytoplasm. These granules usually occur in aggregates just outside the Nissl zone. In addition, aggregates are often present at the extreme periphery of the cells. Most of these secretory granules

are discharged from the perikaryon to the tissue fluid. Some other granules are sent into the axons. The direct relation between the granules and the nucleus is never observed in *Rivulogammarus syriacus*. The nucleolus and other chromatin like basophilic elements are present in the oval nuclei (7–10 μ in diameter) of these cells. The single nucleolus is large, basophilic and invariably found in the peripheral region of the nucleus. These cells are found in all the regions of the nervous system except in dorso-lateral and in the ventral nerve chain groups.

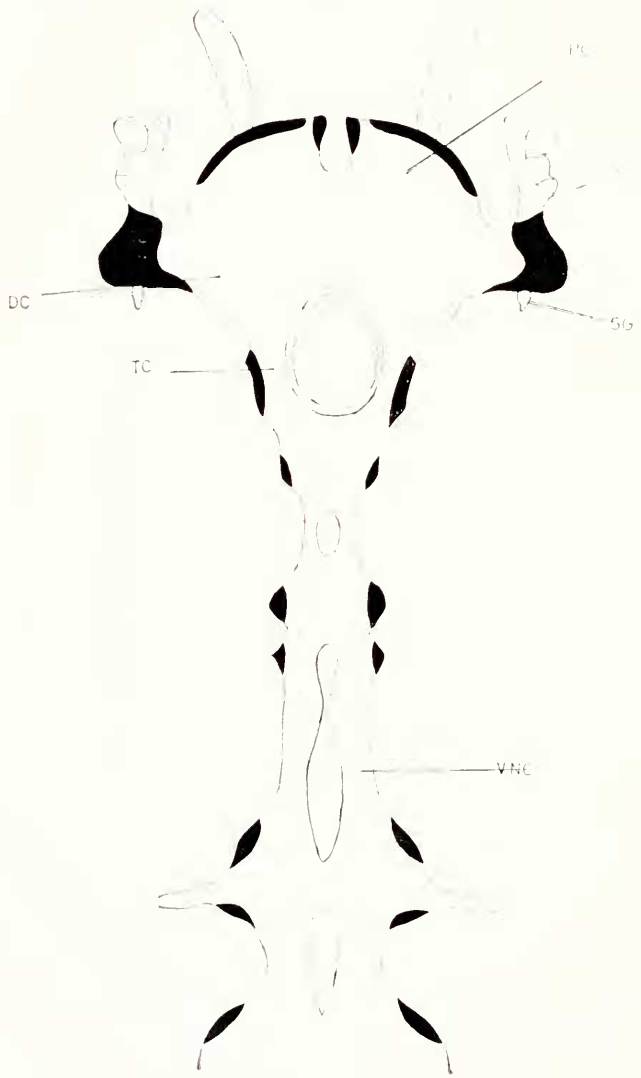
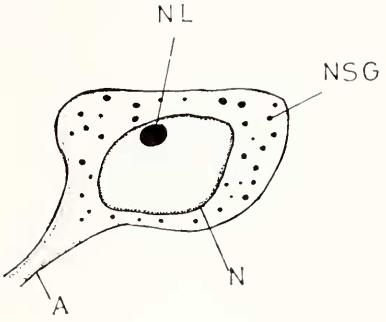


FIGURE 1. Dorsal plan of the central nervous system of *Rivulogammarus syriacus* Chevreux: Black areas show position of neurosecretory cells; PC—Protocerebrum; DC—Deutocerebrum; TC—Tritocerebrum; E—Eye; SG—Sinus gland; VNC—Ventral nerve cord.

B cell (*Figs. 4 and 9*). B cells, about 15 μ in diameter, are oval in shape and are characterized by the extensive granulation of the cytoplasm. The dark blue granules (CHP-positive and PF-negative) are scattered in the cytoplasm and



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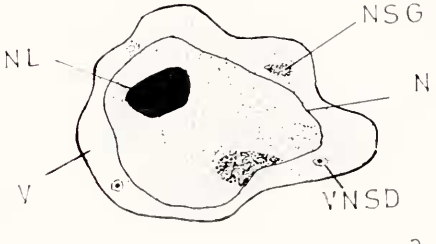

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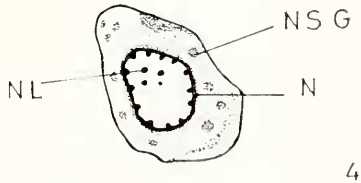

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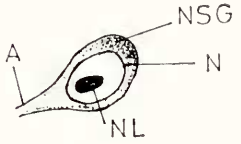

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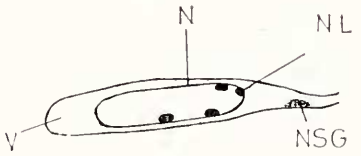

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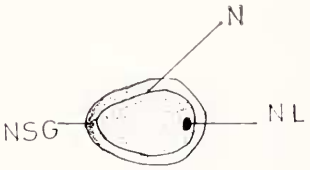

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FIGURE 2. Camera lucida drawing of A type of neurosecretory cell of *Rivulogammarus syriacus* Chevreux; 12X100; A—Axon; N—Nucleus; NL—Nucleolus; NSG—Neurosecretory granules.

FIGURE 3. Camera lucida drawing of B′ type of neurosecretory cell of *Rivulogammarus syriacus* Chevreux; 12X100; N—Nucleus; NL—Nucleolus; NSG—Neurosecretory granules; V—Vacuole; VNSD—Vacuole containing neurosecretory droplet.

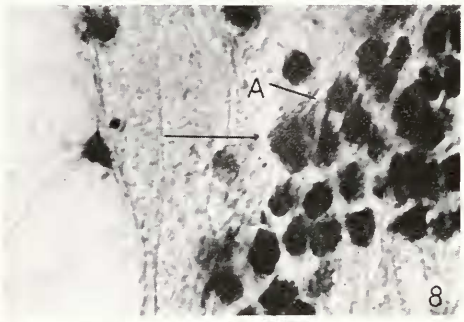
FIGURE 4. Camera lucida drawing of B type of neurosecretory cell of *Rivulogammarus syriacus* Chevreux; 12X100; N—Nucleus; NL—Nucleolus; NSG—Neurosecretory granules.

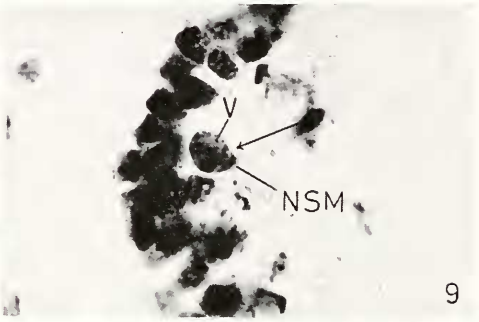
FIGURE 5. Camera lucida drawing of C type of neurosecretory cell of *Rivulogammarus syriacus* Chevreux; 12X100; A—Axon; N—Nucleus; NL—Nucleolus; NSG—Neurosecretory granules.

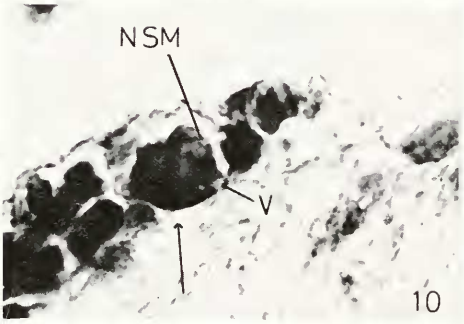
FIGURE 6. Camera lucida drawing of D type of neurosecretory cell of *Rivulogammarus syriacus* Chevreux; 12X100; N—Nucleus; NL—Nucleolus; NSG—Neurosecretory granules; V—Vacuole.

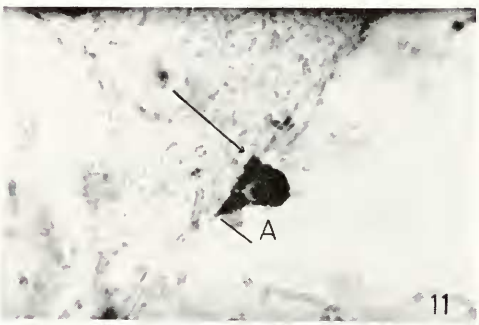
FIGURE 7. Camera lucida drawing of E type of neurosecretory cell of *Rivulogammarus syriacus* Chevreux; 12X100; N—Nucleus; NL—Nucleolus; NSG—Neurosecretory granules.

sometimes they are gathered at one side of the cell and form one or two relatively large masses. Frequently, the large mass of granules disappear from the cell and leaves behind a large vacuole. In most of these cells, the granules form a ring









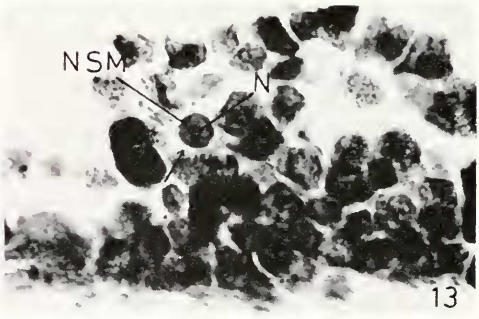


FIGURE 8. Horizontal section through the brain of *Rivulogammarus syriacus* Chevreux, Bouin, CHP. Note A type of cell (arrow), A—Axon.

FIGURE 9. Horizontal section through the brain of *Rivulogammarus syriacus* Chevreux, Bouin, CHP. Note B type of cell (arrow); NSM—Neurosecretory material; V—Vacuole.

FIGURE 10. Horizontal section through the brain of *Rivulogammarus syriacus* Chevreux, Bouin, CHP. Note B′ type of cell (arrow); NSM—Neurosecretory material; V—Vacuole.

FIGURE 11. Horizontal section through the brain of *Rivulogammarus syriacus*, Bouin, CHP. Note C type of cell (arrow); A—Axon.

FIGURE 12. Horizontal section through the brain of *Rivulogammarus syriacus* Chevreux, Bouin, CHP. Note D type of cell (arrow); A—Axon; N—Nucleus.

FIGURE 13. Horizontal section through the brain of *Rivulogammarus syriacus* Chevreux, Bouin, CHP. Note E type of cell (arrow); N—Nucleus; NSM—Neurosecretory material.

outside the nucleus in the vicinity of the Nissl zone. The secretory products are discharged into the tissue fluid directly from the periphery of the cell. The nucleus is distended spherically and measures 5 μ in diameter. It has a densely staining basophilic membrane. The nucleus consists of many peripheral nucleoli lying against the nuclear membrane. The cells are found in all the regions of the nervous system.

B′ cell (*Figs. 3 and 10*). B′ cells are large, 20–25 μ in diameter, and possess little vacuolated cytoplasm. These cells have irregular outlines which are likely caused by shrinkage during fixation. The dense deep stained cytoplasm contains many vacuoles especially at the periphery. The vacuole formation does not have any intimate correlation with nuclear modification. The contents of the vacuoles are found to be CHP- and PF-positive and are discharged directly into the tissue fluid. The empty vacuoles are found in the peripheral region of the cytoplasm. The distended spherical nucleus (12 μ in diameter) has a large basophilic nucleolus and acidophilic nucleoplasm. These cells are found only in the medio-dorsal region of the brain.

C cell (*Figs. 5 and 11*). C cells are small with thick axons and measure about 6–10 μ in diameter. They possess round nuclei 3–6 μ in diameter. The cytoplasm lacks vacuolation and presents a coarse appearance. Irregular masses of dark granules (CHP- and PF-positive) lie scattered around the nucleus. These granules have only one nucleolus. These cells are found in all the groups of the nervous system.

D cell (*Figs. 6 and 12*). D cells are cylindrical, measure 15–20 μ in diameter and have cylindrical nuclei, 8–10 μ in diameter. The deep stained cytoplasm contains a few vacuoles at the periphery. The contents of the vacuoles are found to be CHP-positive and PF-negative. The empty vacuoles are found to be arranged in form of a star. The nucleus contains several nucleoli. These cells are found only in medio-dorsal, antero-lateral and antero-ventral groups of the nervous system.

E cell (*Figs. 7 and 13*). E cells are oval in shape, 8–10 μ in diameter and possess little cytoplasm. The dark blue granules (CHP-positive and PF-negative) are scattered in the cytoplasm and sometimes are found in masses. Furthermore, all the cells do not show the presence of large amount of secretory material at a time. Apparently some cells are at the peak of their secretory process while others are devoid of granules and are in resting stage. The nucleus is round or oval (5–8 μ in diameter) and contains one nucleolus. These cells are found in all the groups of the nervous system.

Neurosecretory cells in the optic lobe

The optic lobe consists of lamina ganglionaris, medulla externa, medulla interna and medulla terminalis. The medulla interna is very poorly developed. There are two large groups of cells found all over the medulla externa and lamina ganglionaris. Since medulla terminalis is not demarcated from the protocerebrum, the neurosecretory cells of both these regions are not differentiated. The neurosecretory cells which are of E type form the anterior and posterior boundaries, 3–4 layers thick (Fig. 14). The X-organ is absent.

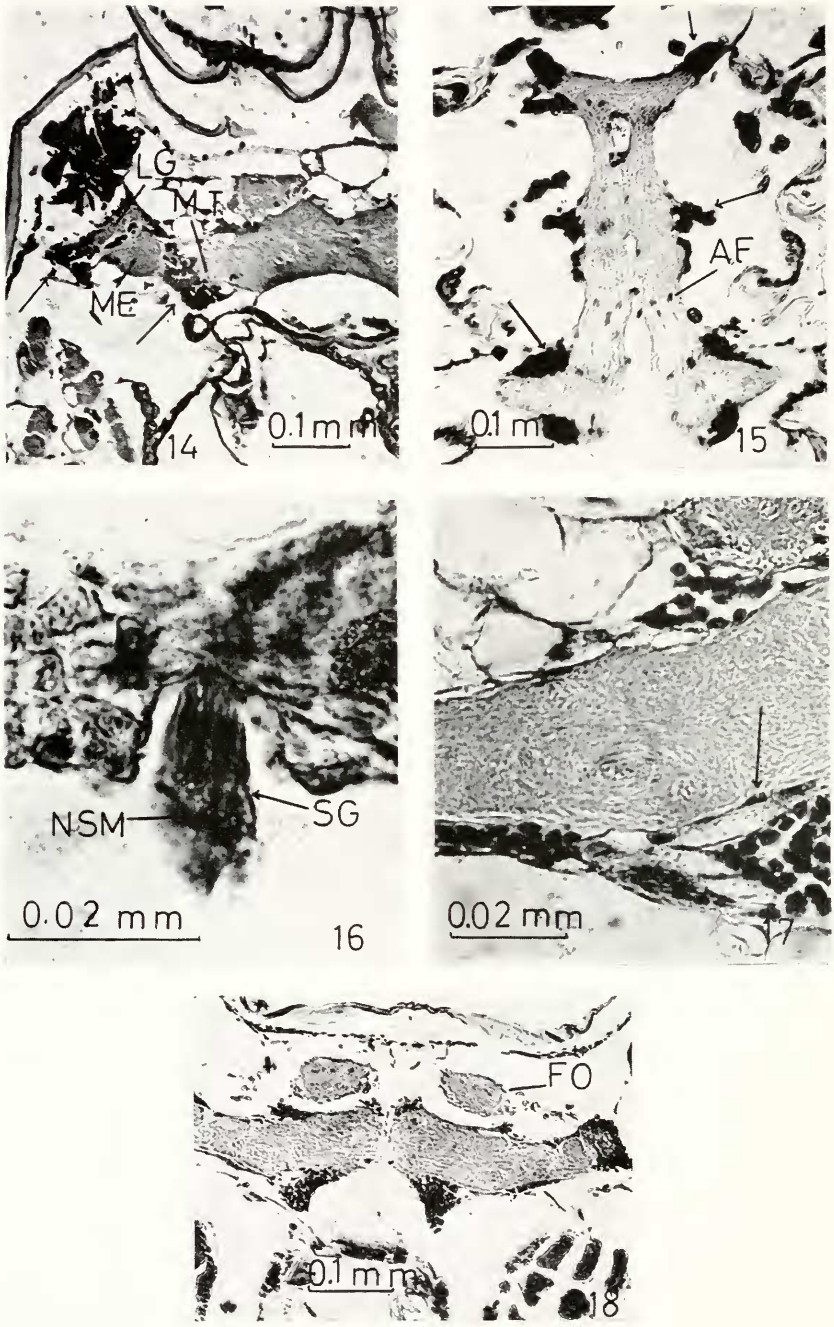


FIGURE 14. Horizontal section through the cephalic region of *Rivulogammarus syriacus* Chevreux showing parts of optic region. Note the various neurosecretory regions (arrows); Bouin, CHP; LG – Lamina ganglionaris; ME—Medulla externa; MT—Medulla terminalis.

Sinus gland

Sinus gland is an oval mass attached by a narrow stalk to the lateral tip and the distal part of the optic lobe. Its smooth capsule is continuous with that of optic lobe, and there are no attachments by blood vessels or connective tissue to any organ except the optic lobe. The sinus gland has a function of storage and release of neurosecretory products, which are elaborated in other sites. It contains nerve endings as does that of decapods, and no nucleus is found in the tissues of the sinus gland. The contents of the sinus gland are stained markedly dark blue with CHP but sometimes acidophilic substances are also detected. With PAS and toluidine blue the contents of the sinus gland are stained pink and violet blue, respectively. It has been observed that the sinus gland has two distinct regions, the distal and proximal (Fig. 16).

Frontal organs

These paired spherical bodies lying anterior to the protocerebrum, are connected to the medulla terminalis of the optic lobe by an oblique nerve. Two kinds of structural elements are observed in the frontal organs. The first kind are the cells without neurosecretory material surrounded by small connective tissue cells and neurons; the second are small round colloidal concretions. The latter seems to be the secretory product of neurosecretory cells and have been transported through the axons (Fig. 18). The neurosecretory material in form of CHP-positive granules are also found along the course of the axons coming from the medulla terminalis of the optic lobe. These neurosecretory materials have PAS-positive reaction.

Neurosecretory activity of different cell types

All the neurosecretory cells fall into two groups on the basis of their neurosecretory activities. The first group is comprised of A, C, and E types of cells. The first stage in the secretory cycle of these cells is marked by the dense granulation of entire cytoplasm and by appearance of small granular bodies within it (Fig. 19). These small granular bodies aggregate into larger and darker ones which are finally discharged either through the cell membrane into the tissue fluid or into the axons. The second group is comprised of B, B′, and D types of cells. The neurosecretory granules form a ring outside the Nissl zone (Fig. 20). The CHP-positive granules in B and D cells and CHP- and PF-positive granules in B′ cells, aggregate to form relatively large masses. Frequently the large masses of granules disappear from the cell and leave vacuoles behind (Fig. 10).

FIGURE 15. Horizontal section through the ventral nerve cord of *Rivulogammarus syriacus* Chevreux, Bouin, CHP. Note the various neurosecretory regions (arrows); AF—Axon fiber carrying neurosecretory material.

FIGURE 16. Horizontal section through cephalic region of *Rivulogammarus syriacus* Chevreux, Bouin, CHP. Note the sinus gland (arrow); NSM—Neurosecretory material; SG—Sinus gland.

FIGURE 17. Horizontal section through the cephalic region of *Rivulogammarus syriacus* Chevreux, Bouin, CHP. Note the axons carrying secretory material (arrow).

FIGURE 18. Horizontal section through cephalic region of *Rivulogammarus syriacus* Chevreux; Bouin, CHP; FO—Frontal organ.

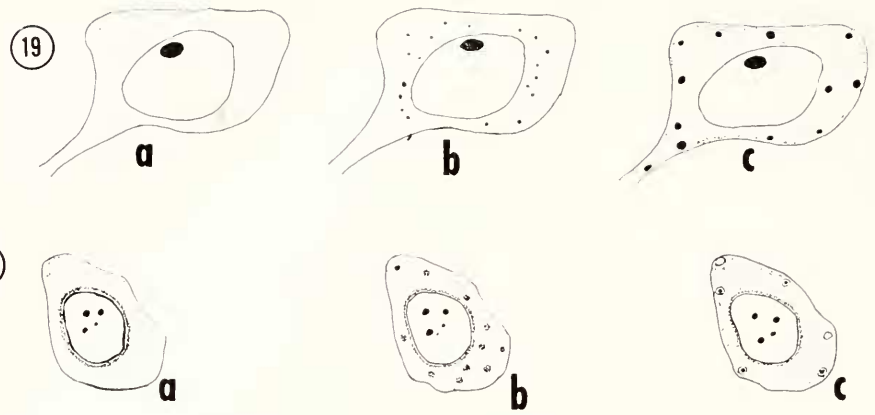


FIGURE 19. Various stages (A, B and C) in the secretory cycle of A type of cell; A—Entire granulation of cytoplasm and appearance of small granular bodies; B—Aggregation of small granular bodies into larger and darker ones; C—Presence of dark bodies in the axon and towards the periphery.

FIGURE 20. Various stages (A, B and C) in the secretory cycle of B type of cell; A—Neurosecretory granules form a ring outside the Nissl zone; B—Aggregation of neurosecretory granules into large masses; C—Release of large neurosecretory masses from the perikaryon and the appearance of vacuoles behind.

The migration of the neurosecretory products of these cells is accomplished in two ways: one is peripheral discharge from the perikaryon as observed in A, B, B′, and E types of cells; the other axonal transportation as seen in A, C, and D types. As is obvious, the migration of the secretory product in A type of cells can take place by both the methods (Fig. 19).

The peripheral discharged substances appear as dark-blue granules into the tissue fluid. These granules migrate with the tissue fluid and are stored in the spaces in a granular form. The axonally transported substances soon loses its identity and aggregate to form large masses. These masses are found in the axon fibers and are carried towards the optic lobes (Fig. 17).

The axon fibers run in the periphery of both anterior and posterior sides of the brain and finally end in the sinus gland. The axons coming from the anterior

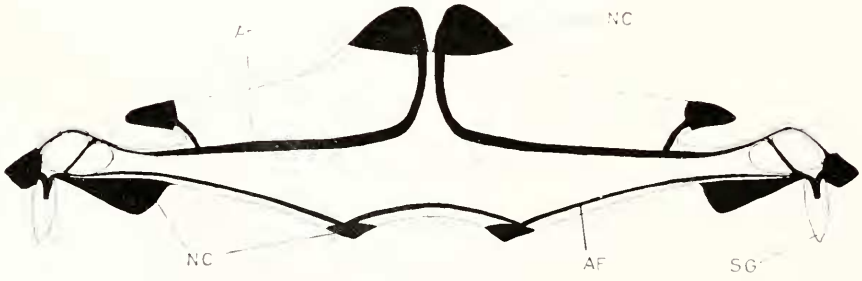


FIGURE 21. Diagrammatic horizontal section of the brain of *Rivulogammarus syriacus* Chevreux showing axons leading towards the sinus gland; AF—Axon fiber; NC—Neurosecretory cells; SG—Sinus gland.

side pass over the medulla externa and then bifurcate into two; one bundle makes a course between the medulla externa and lamina ganglionaris and finally ends in the sinus gland, the other passes over the lamina ganglionaris and then to the sinus gland (Fig. 21).

The neurosecretory axons in the ventral nerve cord can be traced by means of their pink color. The axons which run straight, carry the neurosecretory masses to the brain via circumesophageal connectives (Fig. 15).

Discussion

The neurosecretory cells of the crustaceans have been classified from a morphological point of view by several workers into many types. Enami (1951) described α, β, and γ types of cells in *Sesarma;* Matsumoto reported A, B, C, and D types of cells in *Eriocheir* (1954) and *Chionoectes* (1956) and Inoue (1957) observed A, A′, B, C, D, and E types of cells in *Pachigrapsus*. Later Matsumoto (1958) considered eleven types of cells in *Potamon, Chionoectes, Neptunus* and *Sesarma* but Miyawaki (1960) classified the neurosecretory cells of the decapod crustacea into three types: the giant, the medium and the small. Matsumoto (1959) reported four kinds of neurosecretory cells (A, B, B′, and Y) in an Isopod, *Armadillidium*. As regards Anostraca, Menon (1962) reported A and B types of cells in *Streptocephalus* sp.; Baid and Ramaswamy (1965) observed A and B cells in *Artemia salina* and Hentschel (1965) described U-1, U-2, and U-3 cells in *Chirocephalus* and *Artemia salina*. Recently Lake (1969) observed small cerebral PF-positive monopolar cells, cerebral bipolar PF-positive cells and medium PF-positive in *Chirocephalus diaphanus*.

A histological examination of the central nervous system of *Rivulogammarus syriacus* shows the presence of six kinds of neurosecretory cells (A, B, B′, C, D, and E types). The size of the cells, the mode of discharge of their secretory product and the nature of the secretory granules have formed the basis of such classification.

The morphological characteristics of these cells except A and E types are quite different from that of decapod crustaceans. A type cells of *Rivulogammarus,* judged from their size, would seem to correspond to A cells of *Potamon* (Matumoto, 1958; Baid, Hafidh and Dabbagh, 1968), Enami's (1951) α cells and A cells of *Pachigrapsus* (Inoue, 1957) but the other morphological characteristics of these two cells are also quite different. The nucleus of A cells of these authors show polymorphic appearance and such a condition is not found in A cells of *Rivulogammarus*. The A cells of *Rivulogammarus* correspond to large cells of *Idotea* (Matumoto, 1958) and to A cells of *Armadillidium* (Matumoto, 1959). The E cells of *Rivulogammarus* are quite similar to B cells of other crustaceans (Enami, 1951; Matumoto, 1954, 1958; Inoue, 1957 and Baid, Hafidh and Dabbagh, 1968) concerning their size, shape and structure and stainability of their granules. They are the commonest neurosecretory cells in crustacea. The characteristics of other kinds of cells (B, B′, C, and D) are somewhat different from those of the similarly named cells in crabs. The D types of cells are very distinct, conspicuous and as far as we know, have not been reported earlier.

In entomostracans, the neurosecretory cells have been reported in Cirripedia by Barnes and Gonor (1958), in Cladocera by Sterba (1957), Angel (1966) and

Parker (1966) and in Anostraca by Menon (1962), Baid and Ramaswamy (1965), Hentschel (1965) and Lake (1969). The neurosecretory cells of *Rivulogammarus* do not correspond to any of the cells reported in entomostracans.

The neurosecretory products elaborated in the cytoplasm of the neurosecretory cells, have been studied morphologically by many authors by the use of usual histological techniques (Gomori-chrome-alum haematoxylin phloxine method) and more specialized methods in both vertebrates and invertebrates. Chrome-haematoxylin stained substances (Bliss, Durand and Welsh, 1954), phloxinophilic substances (Matsumoto, 1958), acid fuschin stained substance (Enami, 1951), PAS-positive material (Miyawaki, 1956, 1958) and many other materials stained by various methods have been reported as neurosecretory products. All the cells in the present study are positive to CHP but are negative to PF except B′ and C cells.

Concerning the distribution of neurosecretory cells, A cells are confined to the brain but B, C, and E cells are found both in the brain and the ventral nerve cord ganglia. In the optic lobe of *Rivulogammarus* only E cells are found while in *Armadillidium* Y cells are present (Matumoto, 1959). It is difficult to say whether E cells of *Rivulogammarus* correspond to Y cells of *Armadillidium* (Isopoda). In crabs many kinds of neurosecretory cells are found in the optic lobe but B cells are confined to the ventral corner of the medulla terminalis. In *Armadillium*, they are situated at the anterior side of the brain and at the base of the optic lobe peduncle. The E cells of *Rivulogammarus*, similar to B cells of other crustaceans are distributed in all the regions including the optic lobe. The location of B cells may be considered to show some resemblance in the different orders of Crustacea.

The discharge of neurosecretory substances was earlier believed to take place in two possible ways (Scharrer and Scharrer, 1945; Matumoto, 1958); to these has been added a third by Matumoto (1958). These possibilities are into the tissue spaces of the ganglion, the pedal nerves or the circumesophageal commissures. The axonal transportation of neurosecretory substances has been confirmed by several investigators in the recent past. These observations have contributed to the evolution of the concept of a neurosecretory system in decapod crustacea parallel to the hypothalamic-hypophyseal system of the vertebrates (Bargmann, 1957) and to the inter-cerebralis-cardiacum-allatum system of insects (Scharrer and Scharrer, 1945; Scharrer, 1952). Enami (1951) suggested an axonal transport of neurosecretory material; Bliss and Welsh (1952) came to the conclusion that the neurosecretory substances produced in various parts of the central nervous system migrate along the nerve fibers to the sinus gland where they are stored and eventually released. Matumoto (1954) reported the release of neurosecretory substances in the network of capillaries which closely surround the neurosecretory cells. In the present study, it has been found that the neurosecretory substances are axonally transported and are stored in tissue spaces. The tracing of the pink axon shows that the neurosecretory products of the ventral nerve chain ganglia are transported mainly to the circumesophageal commissure. Further, they are carried to the brain and then to the eyestalks. A similar observation has been reported by Bliss, Durand and Welsh (1954).

The sinus gland in *Gammarus* was described by Graber (1933) as pseudo-frontal organ and by Stahl (1938). Oguro (1959a, 1959b) reported two pairs of sinus glands in three idotead species and in (1960) described a sinus gland in

addition to a pseudofrontal organ in *Tecticeps*. Walker (1936) reported that the pseudofrontal organ described might be identical with the white organ (Koller, 1930) and haemal gland (Hanstrom, 1933) both of which are now designated as sinus gland. Gabe (1952) considered that the pseudofrontal organ is identical with the sinus gland in *Oniscus asellus*. Amar (1950, 1953) stated that the pseudofrontal organ in isopods may be homologous with the sinus gland. Recently, Fingerman (1956) described that with respect to location and structure, the sinus gland of *Ligia exotica* is similar to the organ described by Walker (1936) for *Oniscus asellus*. In *Rivulogammarus* the sinus gland is found to be identical in position and structure to the pseudofrontal organ described by Graber (1933) in *Gammarus*. The sinus gland of this amphipod has two distinct regions: the proximal and the distal. The occurrence of distinct zones, with different tinctorial qualities in the sinus glands of malacostracans has been reported by Enami (1951); Potter (1958) and Carlisle (1959).

There has been a considerable amount of discussion concerning the nomenclature and function of the organs of Bellonci and frontal organs. Claus (1886) described the dorsal frontal organs and regarded them as sensory structures. Hanstrom (1931, 1934) described the frontal organs in *Tanymastix stagnalis* L. and *Polyartemia forcipata* and later (1937), attributed a secretory function to them and regarded them to be precursors of the X-organs of malacostracans. Elofsson (1963) working with different decapods, was able to show that the dorsal frontal organs when present, are always part of the nauplius eye center of the brain. The organs of Bellonci (sensory pore X-organs) are associated with the medulla terminalis in the Decapoda (Knowles and Carlisle, 1956) and have been shown by Dahl (1957) to be derived from neuroblasts of the medulla terminalis. Dahl (1959) has concluded that the dorsal frontal organs and the organs of Bellonci represent phylogenetically quite independent structures because the dorsal frontal organs in the crustacea investigated so far, when present, are associated with the nauplius eye center, whilst the organs of Bellonci are associated with the medulla terminalis. Menon (1962), Elofsson (1966) and Lake (1969) have attributed neurosecretory function to the organs of Bellonci (dorsal front organs) in Anostraca.

The paired frontal organs have been described by Thore (1932) and Hanstrom (1947) in *Gammarus* and with reservations in *Caprella*. According to Graber (1933), these organs do not exist in *Gammarus* and thus those described by others constitute something else than frontal organs. In *Rivulogammarus syriacus* the paired frontal organs are quite distinct and are not innervated from the nauplius eye center but from the medulla terminalis. These paired structures and their axons contain appreciable amount of CHP-positive granules. It appears that these structures are associated with both neurosecretory release and perhaps neurosecretory material synthesis. These functions are similar to those of organs of Bellonci of other malacostracans, the frontal organs of the Copepoda (Carlisle and Pitman, 1961), the X-organs of the Copepoda (Elofsson, 1966), the sensory papilla X-organ of cirriped larvae (Kauri, 1966), and finally to the dorsal frontal organs or X-organs of the Anostraca (Hanstrom, 1934; Menon, 1962 and Elofsson, 1966).

The physiological significance of these six types of neurosecretory cells in central nervous system of *Rivulogammarus syriacus* is yet unknown. As reported in other crustaceans, possibly some of the neurosecretory cells may also be the

source of chromatophorotropic principle. It will be interesting to investigate the physiological significance of the different neurosecretory cells in *Rivulogammarus syriacus*.

Summary

1. The different types of neurosecretory cells occurring in the central nervous system of the freshwater amphipod, *Rivulogammarus syriacus*, their distribution, neurosecretory activity, and the mode of discharge of the neurosecretory substances were studied.

2. There are six types of neurosecretory cells designated as A, B, B′, C, D and E which show definite localization in the central nervous system. The B′ cells are found only in the medio-dorsal region.

3. These cells can be divided into two groups on the basis of their secretory activities.

(a) The first group of cells (A, C and E types) have dense granulated cytoplasm marked by small granular bodies. These bodies aggregate into larger and darker stained ones and are finally discharged either through the cell membrane into the tissue fluid or into the axons.

(b) The second group of cells (B, B′ and D types) have neurosecretory granules in the form of a ring outside the Nissl zone. The granules aggregate to form large masses which disappear through the cell and leave vacuoles behind.

4. The contents of the sinus gland show CHP-positive, PAS-positive, toluidine blue positive and sometimes acidophilic substances.

5. It appears that the frontal organs are associated with neurosecretory release.

6. The physiological activities of these neurosecretory substances in the central nervous system of *Rivulogammarus* are as yet unknown.

Literature Cited

Amar, R., 1950. Les formations endocrines cerebrales des Isopodes marins. *C. R. Acad. Sci. Paris*, **230**: 407–409.

Amar, R., 1951. Formations endocrines cerebrales des Isopodes marins et comportement chromatique d'Idotea. *Ann. Fac. Sci. Marseille*, **20**: 167–306.

Amar, R., 1953. Sur l'existence de cellules neurosecretrices dans le cerveau de *Rocinela*. *Bull. Soc. Zool. Fr.*, **78**: 171–173.

Angel, M. V., 1966. A histological and experimental approach to neurosecretion in *Daphnia magna*. Pages 229–237 in F. Stutinsky Ed., *Neurosecretion*. Fourth International Symposium Neurosecretion. Springer, Berlin.

Baid, I. C., R. A. Hafidh and S. A. Dabbagh, 1968. Studies on the neurosecretory cells of the thoracic ganglion of *Potamon magnum magnum* (Pretzman). *Zool. Anz.*, **180**: 22–32.

Baid, I. C., and L. S. Ramaswamy, 1965. Neurosecretory cells in *Artemia salina* L. *Experientia*, **21**: 528.

Bargmann, W., 1949. Uber die neurosekretorische Verknupfung von Hypothalamus und Neurohypophyse. *Z. Zellforsch.*, **34**: 610–634.

Bargmann, W., 1957. Relationship between neurohypophysial structure and function. Pages 11–22 in H. Heller, Eds., *The Neurohypophysis*. Butterworth, London.

Barnes, H., and J. J. Gonor, 1958. Neurosecretory cells in the cirriped, *Polycipes polymerus*. *J. Mar. Res.*, **17**: 81–102.

Bliss, D. E., J. B. Durand and J. H. Welsh, 1954. Neurosecretory systems in decapod Crustacea. *Z. Zellforsch.*, **39**: 520–536.

Bliss, D. E., and J. H. Welsh, 1952. The neurosecretory system of brachyuran Crustacea. *Biol. Bull.*, **103**: 157–169.

Brown, F. A., Jr., 1944. Hormones in the crustacea: Their sources and activities. *Quart. Rev. Biol.*, **19**: 118–143.

Carlisle, D. B., 1959. On the sexual biology of *Pandalus borealis* (Crustacea: Decapoda). I. Histology of incretory elements. *J. Mar. Biol. Ass. U. K.*, **38**: 381–394.

Carlisle, D. B., and M. J. Pitman, 1961. Diapause, neurosecretion and hormones in Copepoda. *Nature*, **190**: 827–828.

Claus, C., 1886. Untersuchungen Uber die Organisation und Entwicklung von Branchipus und *Artemia* nebst Vegleichenden Bemerkungen Uber andere Phyllopoden. *Arb. Zool. Inst. Univ. Wien*, **6**: 1–104.

Dahl, E., 1957. Embryology of X-organs in *Crangon allmani*. *Nature*, **179**: 482.

Dahl, E., 1963. Main evolutionary lines among recent Crustacea. Pages 1–26 in H. B. Whittington, Ed., *Phylogeny and Evolution of Crustacea*. Museum of Comparative Zoology, special publication.

Dahl, E., 1965. Frontal organs and protocerebral neurosecretory systems in Crustacea and Insecta. *Gen. Comp. Endocrinol.*, **5**: 614–617.

Delattin, 1939. Untersuchungen an Isopode naugen. *Zool. Jb. Anat.*, **1939**: 65.

Elofsson, R., 1963. The nauplius eye and frontal organs in Decapoda (Crustacea). *Sarsia*, **12**: 1–68.

Elofsson, R., 1966. The nauplius eye and frontal organs of the non-Malacostraca (Crustacea). *Sarsia*, **25**: 1–128.

Enami, M., 1951. The sources and activities of two chromatophorotropic hormones in crabs of the genus *Sesarma*. II. Histology of incretory elements. *Biol. Bull.*, **101**: 241–258.

Fingerman, M., 1956. The physiology of the melanophores of the isopod, *Ligia exotica*. *Tulane Stud. Zool.*, **3**: 139–148.

Gabe, M., 1952. Sur l'existence d'un cycle secretoire dans l'organe pseudofrontal (glande du sinus) chez *Oniscus asellus*. *C. R. Acad. Sci. Paris*, **235**: 900–903.

Gabe, M., 1953. Quelques applications de la coloration par la fuchsine-paraldehyde. *Bull. Microsc. Appl.*, **3**: 153–162.

Graber, H., 1933. Uber die Gahirne der Amphipoden und Isopoden. *Z. Morphol. Okol. Tiere*, **25**: 334–371.

Hanstrom, B., 1928. *Vergleichende Anatomie des Nervensystems der wirbellosen Tiere*. Springer, Berlin, 628 pages.

Hanstrom, B., 1929. Das Deutocerebrum der Crustaceen. *Zool. Jahrb. Anat.*, **51**: 535–548.

Hanstrom, B., 1931. Neue Untersuchungen Uber Sinnesorgane und Nervensystem der Crustaceen I. *Z. Morphol. Tiere*, **23**: 80–236.

Hanstrom, B., 1933. Neue Untersuchungen Uber Sinnesorgane und Nervensystem der Crustaceen II. *Zool. Jahrb. Anat.*, **56**: 387–520.

Hanstrom, B., 1934. Neue Untersuchungen Uber Sinnesorgane und Nervensystem der Crustaceen, III. *Zool. Jahrb. Anat.*, **58**: 101–144.

Hanstrom, B., 1937. Inkretorische Organe und Normalfunktionen der Wirbellosen. *Ergebn. Biol.*, **14**: 143–224.

Hanstrom, B., 1947. The brain, the sense organs and the incretory organs of the head in the Crustacea Malacostraca. *Lunds Univ. Arsskr. N. F. Adv.*, **43**: 1–45.

Hentschel, E., 1965. Neurosekretion und Neurohaemalorgan bei *Chirocephalus grubei* Dyb. und *Artemia salina* Leach (Anostraca: Crustacea). *Wiss. Zool.*, **171**: 44–79.

Hubschman, J. H., 1962. A simplified Azan process well studied for crustacean tissue. *Stain Technology*, **37**: 379–380.

Inoue, H., 1957. On the neurosecretory cells of *Pachigrapsus crassipes*. *Memoir Gakugei Faculty Akita University Natural Science*, **70**: 84–92.

Kauri, T., 1966. On the sensory papilla X-organ in Cirriped larvae. *Crustaceana*, **11**: 115–122.

Knowles, F. G. W., and D. B. Carlisle, 1956. Endocrine control in the crustacea. *Biol. Rev.*, **31**: 396–473.

Koller, G., 1928. Versuche uber die inkratorischen Vorgange beim Garneelenfarbwechsel. *Z. Vergl. Physiol.*, **8**: 601–612.

Koller, G., 1930. Weitere Untersuchungen uber Farbwechsel und Farbwechselhormone bei *Crangon vulgaris*. *Z. Vergl. Physiol.*, **12**: 632–667.

Lake, P. S., 1969. Neurosecretion in *Chirocephalus diaphanus* Prevost (Anostraca). I. Anatomy and cytology of the neurosecretory system. *Crustaceana,* **16**: 273–287.

Matsumoto, K., 1954. Neurosecretion in the thoracic ganglion of the crab *Eriocheir japonicus. Biol. Bull.,* **106**: 60–68.

Matsumoto, K., 1956. Migration of the neurosecretion products in the thoracic ganglion of the crab, *Chionoectes opilio. Biol. J. Okayama Univ.,* **2**: 137–146.

Matsumoto, K., 1958. Morphological studies on the neurosecretion in crabs. *Biol. J. Okayama Univ.,* **4**: 103–176.

Matsumoto, K., 1959. Neurosecretory cells of an Isopod, *Armadillidium vulgare* Latreille. *Biol. J. Okayama Univ.,* **5**: 43–50.

Menon, M., 1962. Neurosecretory system of *Streptocephalus* sp. (Anostraca: Branchiopoda). Pages 411–414 in H. Heller and R. B. Clark, Eds., *Neurosecretion.* Academic Press, New York.

Miyawaki, M., 1956. Histological observations on the incretory elements in the eyestalk of a brachyuran, *Telmesus cheirargus* (Tilesius). *J. Fac. Sci. Hokkaido Univ.,* **12**: 325–332.

Miyawaki, M., 1958. On the neurosecretory system of the isopod, *Idotea japonica. Annot. Zool. Jap.,* **31**: 216–221.

Miyawaki, M., 1960. On the neurosecretory cells of some decapod Crustacea. *Kumamoto J. Sci. Series B,* **5**: 1–20.

Oguro, Ch., 1959a. Occurrence of an accessory sinus gland in the isopod, *Idotea japonica. Annot. Zool. Jap.,* **32**: 71–73.

Oguro, Ch., 1959b. On the sinus gland in four species belonging to the Idoteidae (Crust. Isopod.). *J. Fac. Sci. Hokkaido Univ.,* **14**: 261–264.

Oguro, Ch., 1960. On the neurosecretory system in the cephalic region of the isopod, *Tecticeps japonicus. J. Endocrinol.,* **7**: 137–145.

Oguro, Ch., 1961. On the neurosecretory system of two parasitic isopods, *Argeia pugettensis* Dana and *Athelges japonicus* Shiino. *Annot. Zool. Jap.,* **34**: 43–48.

Parker, R. S., 1966. The influence of photoperiod on reproduction and molting of *Daphnia schodleri* Sars. *Physiol. Zool.,* **39**: 266–279.

Passano, L. M., 1951. The X-organ-sinus-gland neurosecretory system in crabs. *Anat. Rec.,* **111**: 502.

Perkins, E. B., 1928. Colour change in crustaceans, especially in *Palaemonetes. J. Exp. Zool.,* **50**: 71–195.

Potter, D. D., 1958. Observations on the neurosecretory system of portunid crabs. Pages 113–118 in W. Bargmann, B. Hanstrom, E. Scharrer and B. Scharrer, Eds., *Neurosekretion.* Second International Symposium, Springer, Berlin.

Scharrer, B., 1952. Neurosekretion. XI. The effect of nerve section on the intercerebralis-cardiacum-allatum system of the insect, *Leucophaea maderae. Biol. Bull.,* **102**: 261–272.

Scharrer, B., 1955. Hormones in invertebrates. Pages 57–95 in G. Pincus and K. V. Thiman, Eds., *The Hormones,* Volume 3. Academic Press, New York.

Scharrer, E., and B. Scharrer, 1945. Neurosecretion. *Physiol. Rev.,* **26**: 171–181.

Stahl, G., 1938. Uber das Vorkommen Von inkretorischen Organen und Farbwechselhormonen im Kopf einiger Crustaceen. *Kungl. Fysiografiska Sallskapers Handlingar. N. F.,* **12**: 1–20.

Sterba, G., 1957. Die neurosekretorischen Zellgruppen einiger Cladoceren (*Daphnia pulex* und *magna, Streptocephalus vetulus*). *Zool. Jahrb. Anat.,* **76**: 303–310.

Thore, S., 1932. Uber statocysten und Frontalorgane bei *Gammarus pulex* und *Gammarus locusta. Zool. Jahrb. Abt.,* **55**.

Walker, R., 1936. The central nervous system of *Oniscus* (Isopoda). *J. Comp. Neurol.,* **62**: 197–238.

Welsh, J. H., 1955. Neurohormones. Pages 97–151 in G. Pincus and K. V. Thimann, Eds., *The Hormones,* Volume 3. Academic Press, New York.